

Rec'd PCT/PTO 12 JAN 2005 #2

P1 1056711

REC'D 02 SEP 2003

WIPO

PCT

# THE UNITED STATES OF AMERICA

**TO ALL TO WHOM THESE PRESENTS SHALL COME:**

UNITED STATES DEPARTMENT OF COMMERCE  
United States Patent and Trademark Office

August 26, 2003

**THIS IS TO CERTIFY THAT ANNEXED HERETO IS A TRUE COPY FROM THE RECORDS OF THE UNITED STATES PATENT AND TRADEMARK OFFICE OF THOSE PAPERS OF THE BELOW IDENTIFIED PATENT APPLICATION THAT MET THE REQUIREMENTS TO BE GRANTED A FILING DATE.**

**APPLICATION NUMBER: 60/397,833**

**FILING DATE: July 23, 2002**

**RELATED PCT APPLICATION NUMBER: PCT/US03/22953**



By Authority of the  
**COMMISSIONER OF PATENTS AND TRADEMARKS**

*M. Sias*

**M. SIAS**  
Certifying Officer

**PRIORITY DOCUMENT**  
SUBMITTED OR TRANSMITTED IN  
COMPLIANCE WITH  
RULE 17.1(a) OR (b)

**BEST AVAILABLE COPY**

07-24-02 13:53 10/23/02

A/Prov

Atty. Dkt. No. 023829-0184

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicants: Shandera, Jr. et al.  
Title: Production of Feedstock With  
Alternative Reducing Agents  
Appl. No.: To be determined  
Filing Date: To be determined  
Examiner: To be determined  
Art Unit: To be determined

<b>CERTIFICATE OF EXPRESS MAILING</b>	
I hereby certify that this correspondence is being deposited with the United States Postal Service's "Express Mail Post Office To Addressee" service under 37 C.F.R. § 1.10 on the date indicated below and is addressed to: Commissioner for Patents, Washington, D.C. 20231	
EV 055956232 US (Express Mail Label Number)	July 23, 2002 (Date of Deposit)
Elizabeth Eiche (Printed Name)	
<i>Elizabeth Eiche</i> (Signature)	

**PROVISIONAL PATENT APPLICATION**  
**TRANSMITTAL**

Commissioner for Patents  
Box PROVISIONAL PATENT APPLICATION  
Washington, D.C. 20231

Sir:

Transmitted herewith for filing under 37 C.F.R. § 1.53(c) is the provisional patent application of:

Donald L. Shandera, Jr.  
640 County Road A  
Ashland, NE 68003

Eugene M. Peters, Jr.  
1589 Constance Avenue  
Kettering, OH 45409

Aharon M. Eyal  
16 Levy  
Jerusalem  
Israel

Ting L. Carlson  
9764 Country Creek Way  
Dayton, OH 45458

Atty. Dkt. No. 023829-0184

## Enclosed are:

- ☒ Specification and Abstract (15 pages) and coversheet (1 page).
- ☒ Application Data Sheet (37 CFR 1.76) (6 pages).
- ☒ Unexecuted Declaration and Power of Attorney (4 sheets)
- ☒ Associate Power of Attorney (2 sheets).

The filing fee is calculated below:

	Rate	Fee Totals
Basic Fee	\$160.00	\$160.00
<input type="checkbox"/> Small Entity Fees Apply (subtract 1/2 of above):	=	\$0.00
<b>TOTAL FILING FEE:</b>	<b>=</b>	<b>\$160.00</b>

- ☒ A check in the amount of \$160.00 to cover the filing fee is enclosed.
- ☒ The Commissioner is hereby authorized to charge any additional fees which may be required regarding this application under 37 C.F.R. §§ 1.16-1.17, or credit any overpayment, to Deposit Account No. 06-1447. Should no proper payment be enclosed herewith, as by a check being in the wrong amount, unsigned, post-dated, otherwise improper or informal or even entirely missing, the Assistant Commissioner is authorized to charge the unpaid amount to Deposit Account No. 06-1447.

Atty. Dkt. No. 023829-0184

Please direct all correspondence to the undersigned attorney or agent at the address indicated below.

Grace P. Malilay, Esq.  
Law Department  
Cargill, Inc.  
P.O. Box 5624  
Minneapolis, MN 55440-5624

Telephone: (952) 742-2561  
Facsimile: (952) 742-6349

Respectfully submitted,

Date

July 23, 2002

FOLEY & LARDNER  
777 East Wisconsin Avenue  
Milwaukee, Wisconsin 53202-5367  
Telephone: (414) 297-5664  
Facsimile: (414) 297-4900

By

Jason E. Pauls

Jason E. Pauls  
Attorney for Applicants  
Registration No. 45,651

**Application Data Sheet****Application Information**

<b>Application Type::</b>	Provisional
<b>Subject Matter::</b>	Utility
<b>Suggested classification::</b>	
<b>Suggested Group Art Unit::</b>	
<b>CD-ROM or CD-R?::</b>	None
<b>Computer Readable Form (CRF)?::</b>	No
<b>Title::</b>	Production of Feedstock With Alternative Reducing Agents
<b>Attorney Docket Number::</b>	023829-0184
<b>Request for Early Publication?::</b>	No
<b>Request for Non-Publication?::</b>	No
<b>Suggested Drawing Figure::</b>	1
<b>Total Drawing Sheets::</b>	0
<b>Small Entity?::</b>	No
<b>Petition included?::</b>	No
<b>Secrecy Order in Parent Appl.?::</b>	No

**Applicant Information**

<b>Applicant Authority Type::</b>	Inventor
<b>Primary Citizenship Country::</b>	US
<b>Status::</b>	Full Capacity
<b>Given Name::</b>	Donald L.
<b>Family Name::</b>	Shandera
<b>Name Suffix::</b>	Jr.
<b>City of Residence::</b>	Ashland
<b>State or Province of Residence::</b>	Nebraska



**Street of mailing address::** 16 Levy  
Jerusalem  
Israel  
**Country of mailing address::** Israel

**Applicant Authority Type::** Inventor  
**Primary Citizenship Country::** US  
**Status::** Full Capacity  
**Given Name::** Ting L.  
**Family Name::** Carlson  
**City of Residence::** Dayton  
**State or Province of Residence::** Ohio  
**Country of Residence::** US  
**Street of mailing address::** 9764 Country Creek Way  
**City of mailing address::** Dayton  
**State or Province of mailing address::** OH  
**Postal or Zip Code of mailing address::** 45458

**Correspondence Information**

**Name::** Grace P. Malilay, Esq.  
**Street of mailing address::** Cargill, Inc.; P.O. Box 5624  
**City of mailing address::** Minneapolis  
**State or Province of mailing address::** Minnesota  
**Postal or Zip Code of mailing address::** 55440-5624  
**Phone number::** (952) 742-2561  
**Fax Number::** (952) 742-6349

**Representative Information**

<b>Representative Designation::</b>	<b>Registration Number::</b>	<b>Representative Name::</b>
Primary	46,521	SCOTT D. ANDERSON
Primary	29,512	RUSSELL J. BARRON
Primary	39,902	DAVID J. BATES
Primary	42,308	STEVEN C. BECKER
Primary	22,022	EDWARD W. BROWN
Primary	35,093	CHARLES G. CARTER
Primary	44,603	ALISTAIR K. CHAN
Primary	26,416	JOHN C. COOPER III
Primary	22,205	WILLIAM J. DICK
Primary	30,844	BARRY L. GROSSMAN
Primary	47,619	JEFFREY S. GUNDERSEN
Primary	44,787	PAUL S. HUNTER
Primary	48,367	JOHN M. LAZARUS
Primary	47,746	KENNETH G. LEMKE
Primary	40,365	KEITH D. LINDENBAUM
Primary	39,282	DAVID G. LUETTGEN
Primary	35,610	RICHARD J. MC KENNA
Primary	32,505	JAMES G. MORROW
Primary	45,651	JASON E. PAULS
Primary	38,276	TODD A. RATHE
Primary	30,128	MICHAEL D. RECHTIN
Primary	48,580	MARCUS W. SPROW
Primary	47,959	M. REED STAHELI
Primary	43,193	JEAN M. TIBBETTS



Primary	44,456	CHRISTOPHER M. TUROSKI
Primary	38,646	JOHN A. VANOPHEM
Primary	34,279	JAMES A. WILKE
Primary	35,421	JOSEPH N. ZIEBERT
Primary	40,883	WALTER E. ZIMMERMAN
Primary	33,647	DAVID L. BIEK
Primary	43,581	PAULA A. DEGRANDIS
Primary	42,512	DANIEL J. ENEBO
Primary	29,000	HARRY J. GWINNELL
Primary	37,669	GRACE P. MALILAY
Primary	42,075	SCOTT T. PIERING
Primary	47,020	PETER M. REYES, JR.
Primary	36,914	PAUL B. SAVEREIDE
Primary	42,152	JEFFREY J. SKELTON

**Domestic Priority Information**

<b>Application::</b>	<b>Continuity Type::</b>	<b>Parent Application::</b>	<b>Parent Filing Date::</b>

**Foreign Priority Information**

<b>Country::</b>	<b>Application number::</b>	<b>Filing Date::</b>	<b>Priority Claimed::</b>

**Assignee Information**

**Assignee name::** Cargill, Inc.

Atty. Dkt. No.: 023829-0184

**U.S. PROVISIONAL PATENT APPLICATION**

**for**

**Production of Feedstock With Alternative Reducing Agents**

**Inventors:** Donald L. Shandera, Jr.  
Eugene M. Peters, Jr.  
Aharon M. Eyal  
Ting L. Carlson

## **Production of Feedstock With Alternative Reducing Agents**

### **BACKGROUND**

**[0001]** The present application relates to cereal processing, and more particularly, to cereal processing using alternative reducing agents.

**[0002]** Traditionally, cereals such as corn, sorghum, wheat, rice and the like have been processed either through wet milling, dry milling or extrusion. Most corn processed in the United States, however, is subject to the wet milling process. This process includes steeping the corn to soften the kernels for separation of the germ, followed by grinding and high-speed centrifugation and or filtration to separate corn germ, protein, fiber and starch. Traditionally, the germ is subsequently processed to vegetable oil, the protein and fiber are used for animal, avian, or fish feed, and the starch is used for either sweetener or alcohol production. The operational costs of wet milling are high because of the energy requirements to dehydrate excess water, operate the high-speed centrifuges to separate desirable matter from undesirable matter, and provide fresh water.

**[0003]** During the traditional steeping process, the cereal material is commonly soaked in an aqueous solution containing a generally recognized as safe (GRAS) reducing agent like sulfur dioxide (SO<sub>2</sub>) to facilitate the reduction and breaking of protein bonds holding together portions of the cereal material. Due to the presence of sulfur dioxide, the steeping liquor is an aqueous solution and works most effectively at a slightly acidic pH. Sulfur dioxide requires replenishing and cannot be easily recycled, and does not effectively reducing many of the proteins of the cereal material.

## SUMMARY

**[0004]** The present application provides a method of processing cereal material using alternative reducing agents. The method includes contacting the cereal material with a solvent to provide softened cereal material. The solvent may be either organic or aqueous, including acids, alkali of metals, ethanol, propanol, glycerol, hexane, acetone, sodium dodecyl sulfate, urea, methysulfoxide, and tetrahydrofuran. The solvent may include at least one reducing agent, with the proviso that at least one reducing agent is not a sulfite, although the solvent desirably does not include sulfites. Alternatively, the reducing agent may be added to the cereal material after the cereal material is contacted with the solvent. The softened cereal material can then be processed to provide at least one cereal component.

**[0005]** As used herein, "cereal material" refers to whole grain or parts derived thereof. Thus, cereal material may include whole grain kernels or portions of kernel at least partially separated by known processes such as dry-milling, wet-milling and/or extrusion. For example, cereal material may include a typical corn kernel having a hull, germ, and endosperm. Cereal material may also include "grits or flour," which refers to material remaining after whole grain kernels have been comminuted and then separated to remove the bulk of the germ material and fibrous outer layer of the kernel, or separated components of cereal including a starch-containing stream, a protein-containing stream, a fiber containing stream, and a germs stream. Common examples of cereal material include corn (maize), sorghum, rice, barley, oats, rye and wheat, but are not limited to such grains.

**[0006]** As used herein, "processing" and "processed" refer to a chemical and/or physical alteration of starch-containing material by one or more operations. Such operations may facilitate penetration of reagents

Atty. Dkt. No.: 023829-0184

or degradation of the cereal material into its component parts. Examples of processing operations may include, without limitation, flaking, steeping, tempering, degerming, dewatering, fiber removal, liquefaction, saccharification, oil extraction, loosening protein coating of starch granules in said cereal kernel, comminuting, degrading, fractionating and/or any operation that facilitates protein separation and/or penetration of at least one agent into the cereal material. The processing operations may be assisted with fermentation products or other chemical and/biological reagents, including operations for hull degradation, liquefaction, saccharification and/or protein degradation of the coating and/or matrix which holds together the starch granules of the cereal material, and fractionating protein, starch, fiber, and germ.

**[0007]** As used herein, "sulfite(s)" refer to sulfur dioxide and the acids and salts of sulfur dioxide when it reacts with water producing a sulfite ion with a reducing potential, including but not limited to sulfurous acid; gaseous sulfur dioxide, or ionic salts of sulfur dioxide such as sodium sulfite, bisodium sulfite, sodium metabisulfite, magnesium sulfite, and ammonium sulfite.

**[0008]** In part, a method is provided to process cereal material to provide a feedstock for non-food applications with the option of using non-GRAS reducing agents. The cereal material may be contacted with a solvent to provide softened cereal material. This may be accomplished by such methods as mixing, immersing, spraying, and misting. The softened cereal material may be further contacted with a reducing agent, as part of the solvent or independent therefrom, with the proviso that the reducing agent does not include sulfites. The softened cereal material may then be processed to provide a feedstock.

**[0009]** In part, a method of processing cereal material is also provided where the cereal material is contacted with a solvent including an effective amount of reducing agent to prevent anti-microbial activity. The

Atty. Dkt. No.: 023829-0184

reducing agent desirably is a non-GRAS reducing agent that is not a sulfite. The cereal material may then be processed to provide cereal material components, which may have independent value (e.g., for use as a feedstock).

[0010] Also provided is a method of processing cereal material by contacting the cereal material with a non-aqueous solvent to provide softened cereal material. Non-exhaustive examples of a non-aqueous solvent may include an alkanol (e.g., ethanol, butanol, glycerol, and propanol), alkane (e.g., hexane and isohexane), ketones (e.g., acetone) or dipolar solvents (e.g., dimethyl sulfoxide and tetrahydrofuran). The solvent may either be a non-sulfite reducing agent or include a non-sulfite reducing agent. The softened cereal material may then be processed to provide a feedstock.

[0011] Potential reducing agents may include mercaptoethanol, dithiothreitol, hydrogen sulfide, glutathione, thioglycolic acid, cysteine, borohydrides, or a mixtures thereof. Additionally, thiol-redox proteins including thioredoxin and glutathione may be used for food grade applications. In all embodiments, the solvent may also include a reductase to allow for regeneration of protein reduction potential. The majority of reducing agent, on a molar equivalent basis, that is used in any one of the above-identified methods is a non-sulfite reducing agent. If sulfites are also present during steeping, they will be present at a level of no more than about 500 ppm, and even more preferably, no more than about 100 ppm.

[0012] It is to be understood that both the foregoing summary and the following detailed description are of a preferred embodiment, and not restrictive of the invention or other alternate embodiments of the invention. A further understanding of the nature and advantages of the information disclosed herein may be realized by reference to the remaining portions of the specification and the drawings. It is to be understood that

the invention is not limited in its application to the details of construction and the arrangements of components set forth in the following description or illustrated in the drawings.

#### DETAILED DESCRIPTION

**[0013]** Provided is the use of alternative reducing agents for use in cereal processing. Generally, cereal material is contacted with a solvent to provide a softened cereal material. The solvent may include an aqueous or non-aqueous solution. The cereal material may be contacted within a holding tank for batch processing or may pass through a system on a continuous basis. Examples of contacting the solvent and cereal material may include, without limitation, mixing, immersing, soaking, spraying or misting.

**[0014]** In one embodiment, the solvent includes a reducing agent, preferably a non-sulfite reducing agent. Non-exhaustive examples of non-sulfite reducing agents include mercaptoethanol, dithiothreitol, hydrogen sulfide, glutathione, thioglycolic acid, cysteine, redox proteins, borohydrides, hypochloride, chlorine, ferrous acid/chloride or any combination thereof. The reducing agents may be used to reduce or break the disulfide bonds that inter-link kernel structural proteins and make the starch susceptible to attack by amylase and/or make starch more readily available for other additional processing such as saccharification. The solvent may also include more than one reducing agent, including sulfites, non-sulfites and combinations thereof. Mixtures of reductants may prove to be more effective than just one; both in reaction rate and concentrations needed. For example, mercaptoethanol and hydrogen sulfide or sulfurous acid, thereby reducing the time required to steep the cereal material. Alternatively, a strong oxidizing agent such as performic acid could be used instead of a reducing agent to break the disulfide bonds of the cereal.



**[0015]** In instances where sulfites are used in addition to non-sulfite reducing agents, the non-sulfite reducing agent should be present at a higher level in final concentration on a molar equivalent basis than the sulfite. Desirably, sulfites are present at a level of no more than about 500 ppm, and more preferably, no more than about 300 ppm, and even more preferably no more than about 100 ppm. Alternatively, the reducing agent (or combination of reducing agents) may be added to the softened cereal material-solvent mixture after the cereal material is contacted with the solvent.

**[0016]** The reducing agents may vary in reducing potential. A number of non-sulfite reducing agents generally have a greater reducing potential than sulfite alone. By using these non-sulfite reducing agents in the processing of cereal material, lower concentrations of reducing agents are required, and steep or tempering time may be reduced, all reducing capital investments and energy costs of the system.

**[0017]** The reducing agents of the present invention may have permanent reactivity in that they completely and permanently react with the disulfide groups of the peptides within the kernel. Once reacted, the reducing agent is not required to be present for further processing. As a result, the quantity of reducing agents used for contacting with the cereal material needs to be matched only to the quantity of cereal material in a desired molecular ratio or in excess of that required. The desired molar ratio may vary from the quantity of disulfide bonds to the quantity of cysteine residues within the grain. For example, in corn the cysteine content varies from approximately 0.15 to 0.20 moles per bushel of corn. The concentrations of reducing agent used within the process may be adjusted to prevent residual reducing agents in the remaining solvent upon separation of the softened cereal material or downstream products.

**[0018]** The processing conditions for contacting the cereal material with solvent are regulated by the reducing agents effective pH range and

Atty. Dkt. No.: 023829-0184

absorption rates. Non-sulfite reducing agents can be used to provide a wide range of processing conditions. For example, cereal material may be contacted with solvent and maintained at pH of 2 when using hypochlorite as a reducing agent, which is outside of current pH processing ranges of 3 to 5.5. Other reducing agents such as dithiothreitol may be used at a neutral pH range of 5.6 to 8, while reducing agents such as mercaptoethanol may be used at alkaline pH range of 8 to 14. Generally, the pH may be in the activity range of the specific reducing agent used in the system.

**[0019]** In other embodiments, non-sulfite reducing agents may allow cereal material to be tempered, steeped, or contacted with alternative solvents. Other conditions that may be altered include the type of solvent, the use of oxygen, and temperature within the system. Non-sulfite reducing agents may be used in organic and alternative aqueous solutions, such as ethanol, urea, dimethylsulfoxide, acetone and hexane, and still retain reductant functionality that sulfur dioxide or other sulfites would lose when in the same or similar solvent. For example, the reducing agent mercaptoethanol may be used with ethanol to soften the cereal material, and at least partially degrade the disulfide bonds of the peptides contained therein. Also, when using non-sulfite reducing agents, oxygen presence or atmospheric exposure of the steeping system may not be required, or if required, at reduced levels, unlike when using sulfites. Further, corn could be steeped at alternative temperatures, thus reducing energy costs.

**[0020]** In an alternative embodiment, thiol-redox proteins may be used as reducing agents. The thio-redox proteins may include proteins such as thioredoxin. These proteins may enzymatically reduce the peptide of the cereal material. For example, gamma-zein is a protein common in the endosperm and may be enzymatically reduced by thioredoxin. To oxidize or recharge the thiol-redox protein, a reductase and a secondary

Atty. Dkt. No.: 023829-0184

compound such as NADPH may be provided to the solvent or moisturized cereal material. This will permit a reusable supply of reducing agent.

**[0021]** Lost biomass may be recovered using non-sulfite reducing agents in processing cereal material. When used at effective levels, non-sulfite reducing agents such as mercaptoethanol and dithiothreitol are powerful reductants that have an (anti)microbial activity. At an effective level, microbial growth is slowed or inhibited, or the reducing agent may cause apoptosis. A reduction in the number of microbes increases the amount of nutrients such as sugars, nucleic acids, vitamins, minerals, peptides and other metabolites that are typically leached from the kernel and fermented or metabolized. For example, *Lactobacillus* is a common bacteria present during the steeping stage of cereal processing, which converts available nutrients to lactic acid. By reducing or inhibiting *Lactobacillus* growth, these nutrients (e.g., biomass) can be recovered (e.g., from the steepwater and sold as media or added to the feedstock).

**[0022]** The softened cereal material may be processed to provide at least one of a starch containing stream and a protein containing stream. The streams contain a higher ratio of the stated materials than impurities (e.g., extraneous materials including water, residual reducing agent, and other cereal material components). At least a portion of the streams may be separated and/or combined and further processed to produce a feedstock.

#### EXAMPLES

**[0023]** The following examples are presented to illustrate the present invention and to assist one of ordinary skill in making and using the same. The examples are not intended in any way to otherwise limit the scope of the invention.

Example 1)

**[0024]** Corn material (whole kernels and/or endosperm pieces) is steeped in a counter current process where solvent containing the greatest concentration of reducing agent is added opposed to the freshest corn. Processing solvent is added at a ratio of about 3 to 12 gal/bu cereal material. Steeping may occur for 1-48 hr, with 3-35 hrs being preferred, dependent on level of comminution induced on starting material. Heating may be done by heating water or direct steam injection on the kernels and or components. Although steeping may be performed at ambient temperature for many reductants, steeping at elevated temperature to maximize reaction rates is preferred with a typical steeping temperature of about 50°C preferred.

**[0025]** The processing solvent is an aqueous solution of dithiothreitol (DTT) to reduce disulfide linkages of endosperm protein for protein denaturing and release of starch granules by a wet process. The reducing agent is added to the aqueous solution in the amounts of 0.1 to 500 millimolar with a preferred concentration of 10 to 100 mM. The optimal concentration is adjusted in a molar ratio equivalent or slightly greater than the molar quantity of disulfide groups within the kernels, but not with excess that residual reductant will contaminate the light steepwater or fermentation feedstock at concentrations of greater than 10 mM after steeping. The steep solution pH is neutral or slightly acidic (5-7) as an effect of the solution used and buffering capacity of the corn, but the solution pH may be optimized for enhancing reducing agents chemical activity and stability in the system.

**[0026]** After steeping, the cereal may be exposed to grinding, attrition or other shearing forces to aid structural degradation of the cereal and separation into primary components, if so desired. Conventional separation processes such as high-speed centrifugation and filtration methods may be employed to remove the bulk of the germ material and fibrous outer layer of the kernel if present. Protein and starch may be

further separated to provide a protein-enriched stream and a starch enriched stream using one or more conventional separation processes such as centrifugation and filtration.

[0027] Fermentation feedstock may be produced from the carbohydrate-enriched stream by conventional methods such as jetting and acid and or enzymatic hydrolysis.

Example 2)

[0028] The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but pH of the steep solution is adjusted to an acidic pH of less than 3 using an acid. A reducing agent with activity at acidic pH, such as hypochlorite, is added to the steep solution at a molar ratio equivalent or greater than the cysteine content of the corn.

Example 3)

[0029] The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but pH of the steep solution is buffered to an alkaline pH of greater than 7 using an alkali of metal with a reductant effective at alkaline pH such as mercaptoethanol. Adjustment of pH above 8.5, pH greater than 10 preferred, increases the rate of kernel softening and steeping effectiveness. Steeping is performed in a manner such as nitrogen blanketing to minimize oxygen content of the steeping environment and potential oxidation and loss of reductants' reduction potential.

Example 4)

[0030] The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but a combination of reductants is used to synergistically maximize the rate and extent of reduction. A reductant with terminal reactivity such as dithiothreitol is combined with a low dosing of hydrogen sulfide or sulfites. The lower molecular weight

reductants have greater penetration capability within the endosperm and somewhat loosen the primary structure of the kernel, allowing the dithiothreitol to penetrate faster and complete the reactions.

Example 5)

[0031] Steeping is performed in an atmospheric oxygen-protected system of tanks or screw conveyor to exclude atmospheric oxygen and prevent oxidation of reducing agents and loss of effectiveness. The system may be further protected with a blanketed inert gas such as nitrogen to further prevent oxidation and loss of reducing agent activity. Additionally, excess reducing agent or sulfites may be added to scavenge free oxygen from the steep solution and corn. Exclusion of oxygen is recommended when steeping at alkaline pH.

Example 6)

[0032] The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but a reducing agent(s) such as mercaptoethanol or dithiothreitol is added in excess within the steep to inhibit microbial growth within the steeping system. Inhibition of microbial growth prevents loss to biomass (typically in fermentation) of sugars, nucleic acids, vitamins, minerals, peptides and other nutrients typically leached from the kernel or otherwise present in the steep solution. These nutrients are recaptured in light steepwater and may be added to the fermentation feedstock as a source of nutrients. Excess reductants may be oxidized to prevent deleterious effects on fermentation application.

Likewise, products of examples 2 and 3 may show inhibition of microbial growth within steeping without addition of excess reductant due to the extreme pH of the systems. Likewise example 1 would have similar effect if steeping is performed at high temperatures (e.g. greater than 60°C).

The steepwater from such processes would likewise be rich in sugars, nucleic acids, vitamins, minerals, peptides, etc. because of the lack of microbial growth and utilization.

Atty. Dkt. No.: 023829-0184

**Example 7)**

**[0033]** The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but in a solution of hexane with a reducing agent(s) such as mercaptoethanol.

**Example 8)**

**[0034]** The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but in a solution of butanol with a reducing agent such as mercaptoethanol added.

**Example 9)**

**[0035]** The corn or corn components (e.g. grits and or flour) are steeped as in example 1, but in a solution of greater than 80% dimethylsulfoxide is used with a reducing agent such as mercaptoethanol added. Dimethylsulfoxide itself degrades the kernel structure and depolymerizes and solubilizes the kernel components. Mercaptoethanol reduces the disulfide bonds to quicken and aid the process.

Mercaptoethanol is more effective in this application than sulfites due to the limited water content of the system. Use of solvents such as dimethylsulfoxide also perform as a process aid in gelatinizing the starch and reducing the amount of energy, chemicals, and or enzyme for conversion of the starch and protein to fermentation feedstock (e.g. reducing or replacing jetting operations).

## ILLUSTRATIVE EMBODIMENTS

**[0036]** Provided is a method of processing cereal material including: contacting the cereal material in solvent to provide softened cereal material; and processing the softened cereal material; wherein the solvent includes at least one reducing agent, with the proviso that at least one reducing agent does not include sulfites.

Atty. Dkt. No.: 023829-0184

# ILLUSTRATIVE EMBODIMENTS

**[0036]** Provided is a method of processing cereal material including: contacting the cereal material in solvent to provide softened cereal material; and processing the softened cereal material; wherein the solvent includes at least one reducing agent, with the proviso that at least one reducing agent does not include sulfites.

**[0037]** The reducing agent may include mercaptoethanol, dithiothreitol, hydrogen sulfide, glutathione, thioglycolic acid, cysteine, redox protein, borohydride, glutathione, thioglycolic acid, hypochloride, chlorine, ferrous acid/chloride or a combination thereof. If sulfite is present, the solvent or softened cereal material should include no more than about 500 ppm sulfites, and more preferably, no more than about 100 ppm sulfites.

**[0038]** The cereal material may be processed to provide at least one of a starch containing stream and a protein containing stream. Other methods may include separating and/or combining the stream from other cereal material components; and processing the streams to produce a feedstock. Examples of processing to produce a feedstock may include at least one of a liquefying operation, a hydrolyzing operation and a saccharifying operation.

**[0039]** The cereal material used may include corn, wheat, barley, rye, oats, sorghum, millet, rice or a combination thereof. The cereal material may include defatted grits, debranned grits, flour or mixtures thereof.

**[0040]** Further, the solvent may be aqueous or organic. The solvent may also have an acidic, a neutral, or an alkaline pH, depending on the effective pH range of the reducing agent. The solvent may also include urea, surfactants, plasticizers, alkali of metals, or a combination thereof.

**[0041]** In one embodiment, the solvent may include a reagent to facilitate replenishment or reactivation of the reduction potential of the



Atty. Dkt. No.: 023829-0184

reducing agent. The reagent may be a reductase when the reducing agent is a thio-redox protein.

**[0042]** The solvent may include the reducing agent, in which case the reducing agent may be contacted with the softened cereal material prior to processing the cereal material. Alternatively, the reducing agent may be contacted with the softened cereal material after processing the cereal material. For example, the reducing agent is contacted with the cereal material after debranning or degerming, prior to shearing or cooking, or after shearing or cooking. Generally, the reducing agent is contacted with the cereal material at a molar ratio to the cereal material, or in excess.

**[0043]** Also provided is a method of processing cereal material including: contacting the cereal material with a solvent to provide softened cereal material; and processing the cereal material to provide a feedstock; wherein the solvent includes an effective amount of reducing agent to prevent anti-microbial activity, with the proviso that the reducing agent does not include sulfites.

**[0044]** In another embodiment, a method of processing cereal material is provided. The method may include: contacting the cereal material with a non-aqueous solvent to provide softened cereal material; and processing the softened cereal material to provide a feedstock; wherein the solvent includes a reducing agent, with the proviso that the reducing agent does not include sulfites.

Atty. Dkt. No.: 023829-0184

## ABSTRACT

The present application provides a method of processing cereal material using alternative reducing agents. The method includes contacting the cereal material with a solvent to provide softened cereal material. The  
5 solvent may include at least one reducing agent, with the proviso that at least one reducing agent is not a sulfite.

**DECLARATION AND POWER OF ATTORNEY**

As a below named inventor, I HEREBY DECLARE:

THAT my residence, post office address, and citizenship are as stated below next to my name;

THAT I believe I am the original, first, and sole inventor (if only one inventor is named below) or an original, first, and joint inventor (if plural inventors are named below or in an attached Declaration) of the subject matter which is claimed and for which a patent is sought on the invention entitled

Production of Feedstock With Alternative Reducing Agents

(Attorney Docket No. 023829-0184)

the specification of which (check one)

  X   is attached hereto.

       was filed on        as United States Application Number or PCT International Application Number        and was amended on        (if applicable).

THAT I do not know and do not believe that the same invention was ever known or used by others in the United States of America, or was patented or described in any printed publication in any country, before I (we) invented it;

THAT I do not know and do not believe that the same invention was patented or described in any printed publication in any country, or in public use or on sale in the United States of America, for more than one year prior to the filing date of this United States application;

THAT I do not know and do not believe that the same invention was first patented or made the subject of an inventor's certificate that issued in any country foreign to the United States of America before the filing date of this United States application if the foreign application was filed by me (us), or by my (our) legal representatives or assigns, more than twelve months (six months for design patents) prior to the filing date of this United States application;

THAT I have reviewed and understand the contents of the above-identified specification, including the claim(s), as amended by any amendment specifically referred to above;

THAT I believe that the above-identified specification contains a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention, and sets forth the best mode contemplated by me of carrying out the invention; and

THAT I acknowledge the duty to disclose to the U.S. Patent and Trademark Office all information known to me to be material to patentability as defined in Title 37, Code of Federal Regulations, §1.56.

Atty. Dkt. No. 023829-0184

I HEREBY CLAIM foreign priority benefits under Title 35, United States Code § 119(a)-(d) or § 365(b) of any foreign application(s) for patent or inventor's certificate, or § 365(a) of any PCT international application which designated at least one country other than the United States of America, listed below and have also identified below any foreign application for patent or inventor's certificate or of any PCT international application having a filing date before that of the application on which priority is claimed.

Prior Foreign Application Number	Country	Foreign Filing Date	Priority Claimed?	Certified Copy Attached?

I HEREBY CLAIM the benefit under Title 35, United States Code § 119(e) of any United States provisional application(s) listed below.

U.S. Provisional Application Number	Filing Date

I HEREBY CLAIM the benefit under Title 35, United States Code, § 120 of any United States application(s), or § 365(c) of any PCT international application designating the United States of America, listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States or PCT International application in the manner provided by the first paragraph of Title 35, United States Code, § 112, I acknowledge the duty to disclose information which is material to patentability as defined in Title 37, Code of Federal Regulations, § 1.56 which became available between the filing date of the prior application and the national or PCT international filing date of this application.

U.S. Parent Application Number	PCT Parent Application Number	Parent Filing Date	Parent Patent Number

I HEREBY APPOINT the following registered attorneys and agents of the law firm of FOLEY & LARDNER:

SCOTT D. ANDERSON  
 RUSSELL J. BARRON  
 DAVID J. BATES  
 STEVEN C. BECKER  
 EDWARD W. BROWN  
 CHARLES G. CARTER  
 ALISTAIR K. CHAN  
 JOHN C. COOPER III

Reg. No. 46,521  
 Reg. No. 29,512  
 Reg. No. 39,902  
 Reg. No. 42,308  
 Reg. No. 22,022  
 Reg. No. 35,093  
 Reg. No. 44,603  
 Reg. No. 26,416

**Atty. Dkt. No. 023829-0184**

WILLIAM J. DICK	Reg. No.	22,205
BARRY L. GROSSMAN	Reg. No.	30,844
JEFFREY S. GUNDERSEN	Reg. No.	47,619
PAUL S. HUNTER	Reg. No.	44,787
JOHN M. LAZARUS	Reg. No.	48,367
KENNETH G. LEMKE	Reg. No.	47,746
KEITH D. LINDENBAUM	Reg. No.	40,365
DAVID G. LUETTGEN	Reg. No.	39,282
RICHARD J. MC KENNA	Reg. No.	35,610
JAMES G. MORROW	Reg. No.	32,505
JASON E. PAULS	Reg. No.	45,651
TODD A. RATHE	Reg. No.	38,276
MICHAEL D. RECHTIN	Reg. No.	30,128
MARCUS W. SPROW	Reg. No.	48,580
M. REED STAHELI	Reg. No.	47,959
JEAN M. TIBBETTS	Reg. No.	43,193
CHRISTOPHER M. TUROSKI	Reg. No.	44,456
JOHN A. VANOPHEM	Reg. No.	38,646
JAMES A. WILKE	Reg. No.	34,279
JOSEPH N. ZIEBERT	Reg. No.	35,421
WALTER E. ZIMMERMAN	Reg. No.	40,883

to have full power to prosecute this application and any continuations, divisions, reissues, and reexaminations thereof, to receive the patent, and to transact all business in the United States Patent and Trademark Office connected therewith.

**I request that all correspondence be directed to:**

**Grace P. Malilay, Esq.**  
**Cargill, Inc.**  
**P.O. Box 5624**  
**Minneapolis, MN 55440-5624**

**Telephone:** (952) 742-2561  
**Facsimile:** (952) 742-6349

**I UNDERSTAND AND AGREE THAT the foregoing attorneys and agents appointed by me to prosecute this application do not personally represent me or my legal interests, but instead represent the interests of the legal owner(s) of the invention described in this application.**

I FURTHER DECLARE THAT all statements made herein of my own knowledge are true, and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Atty. Dkt. No. 023829-0184

Name of first inventor	Donald L. Shandera, Jr.
Residence	Ashland, Nebraska
Citizenship	US
Post Office Address	640 County Road A Ashland, NE 68003
Inventor's signature	
Date	
Name of second inventor	Eugene M. Peters, Jr.
Residence	Kettering, Ohio
Citizenship	US
Post Office Address	1589 Constance Avenue Kettering, OH 45409
Inventor's signature	
Date	
Name of third inventor	Aharon M. Eyal
Residence	Jerusalem, Israel
Citizenship	Israel
Post Office Address	16 Levy Jerusalem Israel Israel
Inventor's signature	
Date	
Name of fourth inventor	Ting L. Carlson
Residence	Dayton, Ohio
Citizenship	US
Post Office Address	9764 Country Creek Way Dayton, OH 45458
Inventor's signature	
Date	

Attorney Docket No. 023829-0184

**IN THE UNITED STATES PATENT AND TRADEMARK OFFICE**

Applicant(s): Shandera, Jr. et al.


Title: Production of  
Feedstock with  
Alternative Reducing  
Agents

Application No.: To be determined

Filing Date: To be determined

Examiner: To be determined

Art Unit: To be determined

CERTIFICATE OF EXPRESS MAILING	
I hereby certify that this correspondence is being deposited with the United States Postal Service's "Express Mail Post Office To Addressee" service under 37 C.F.R. § 1.10 on the date indicated below and is addressed to: U.S. Commissioner of Patents and Trademarks, Washington, D.C. 20231.	
EV 055956232 US (Express Mail Label Number)	July 23, 2002 (Date of Deposit)
Elizabeth Eiche (Printed Name)	
 (Signature)	

U.S. Commissioner of Patents and Trademarks  
Washington, D.C. 20231

**ASSOCIATE POWER OF ATTORNEY**

The undersigned attorney of record in the above-identified patent application hereby appoints the following registered attorneys of Cargill, Inc., a corporation duly organized and existing under the laws of the State of Delaware, and having a place of business at 15407 McGinty Road W., Wayzata, Minnesota, as associate attorneys in said application to have full power to prosecute this application and any continuations, divisions, reexaminations, and reissues thereof, to receive any issued patent, and to transact all business in the United States Patent and Trademark Office connected therewith:

DAVID L. BIEK  
PAULA A. DEGRANDIS  
DANIEL J. ENEBO  
HARRY J. GWINNELL  
GRACE P. MALILAY  
SCOTT T. PIERING  
PETER M. REYES, JR.  
PAUL B. SAVEREIDE  
JEFFREY J. SKELTON

Reg. No. 33,647  
Reg. No. 43,581  
Reg. No. 42,512  
Reg. No. 29,000  
Reg. No. 37,669  
Reg. No. 42,075  
Reg. No. 47,020  
Reg. No. 36,914  
Reg. No. 42,152

Attorney Docket No. 023829-0184

Please direct all future correspondence concerning this application to:

Grace P. Malilay, Esq.  
Law Department  
Cargill, Inc.  
P.O. Box 5624  
Minneapolis, Minnesota 55440-5624

Registration No.: 37,669  
Telephone: (952) 742-2561  
Facsimile: (952) 742-6349

Respectfully submitted,

Date July 23, 2002

FOLEY & LARDNER  
777 East Wisconsin Avenue  
Suite 3800  
Milwaukee, Wisconsin 53202-5367  
Telephone: (414) 297-5664  
Facsimile: (414) 297-4900

By Jason E. Pauls

Jason E. Pauls  
Attorney for Applicant(s)  
Registration No. 45,651



**This Page is Inserted by IFW Indexing and Scanning  
Operations and is not part of the Official Record**

**BEST AVAILABLE IMAGES**

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

- ☒ **BLACK BORDERS**
- ☐ **IMAGE CUT OFF AT TOP, BOTTOM OR SIDES**
- ☒ **FADED TEXT OR DRAWING**
- ☒ **BLURRED OR ILLEGIBLE TEXT OR DRAWING**
- ☐ **SKEWED/SLANTED IMAGES**
- ☐ **COLOR OR BLACK AND WHITE PHOTOGRAPHS**
- ☐ **GRAY SCALE DOCUMENTS**
- ☐ **LINES OR MARKS ON ORIGINAL DOCUMENT**
- ☒ **REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY**
- ☐ **OTHER:** \_\_\_\_\_

**IMAGES ARE BEST AVAILABLE COPY.**

**As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.**